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- (71) Applicant (for all designated States except US): XY, INC. [US/US]: 1108 North Lemay Avenue, Fort Collins, CO 80524 (US).
- (72) Inventors; and
- (75) Inventors/Applicants (for US only): EVANS, Kenneth, M. [US/US]: 2125 Pinrose Lane, Fort Collins, CO 80526 (US). VAN MUNSTER, Erik, B. [NL/US]: Lijnbaansgracht 28C, NL-1015 GP Amsterdam (NL).
- (74) Agent: MILES, Craig, R.; Santangelo Law Offices, P.C., Third Floor, 125 South Howes, Fort Collins, CO 80521 (US).
- (81) Designated States (national): AE, AG, AL, AM, AT, AT (utility model), AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, CZ (utility model), DE, DE (utility model), DK, DK (utility model), DM, DZ, EE, EE (utility model), ES, FI, FI (utility model), GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SK (utility model), SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZW.
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- Declaration under Rule 4.17:**  
— of inventorship (Rule 4.17(iv)) for US only
- Published:**  
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- For two-letter codes and other abbreviations, refer to the "Guidance Notes on Codes and Abbreviations" appearing at the beginning of each regular issue of the PCT Gazette.

(54) Title: HIGH PURITY X-CHROMOSOME BEARING AND Y-CHROMOSOME BEARING POPULATIONS OF SPERMATOZOA

(57) Abstract: Isolated non-naturally occurring populations of spermatozoa (15) having high purity and technologies to differentiate spermatozoa (28) based on characteristics such as mass, volume, orientation, or emitted light including methods of analysis and apparatus such as beam shaping optics (30) and detectors (32).

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## INTERNATIONAL SEARCH REPORT

International application No.

PCT/US01/15150

**A. CLASSIFICATION OF SUBJECT MATTER**

IPC(7) : A01N 1/02; C12M 3/00  
 US CL : 435/2, 288.7

According to International Patent Classification (IPC) or to both national classification and IPC

**B. FIELDS SEARCHED**

Minimum documentation searched (classification system followed by classification symbols)

U.S. : 435/2, 288.7

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched  
NONEElectronic data base consulted during the international search (name of data base and, where practicable, search terms used)  
USPATFULL on West, WPIDS, AGRICOLA on STN**C. DOCUMENTS CONSIDERED TO BE RELEVANT**

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	BEYHAN et al. Sexual dimorphism in IVM-IVF bovine embryos produced from X and & chromosome-bearing spermatozoa sorted by high speed flow cytometry. 01 July 1999, Vol. 52, No. 1, pages 35-48, especially abstract.	1-11, 13-22, 25-29
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Y		30, 31
A.P	US 6,154,276 A (MARIELLA, Jr.) 28 November 2000.	1-50, 96-209
A	US 5,985,216 A (RENS et al.) 16 November 1999.	1-50, 96-209
A.P	WO 01/51612 A1 (GALLI et al.) 19 July 2001.	1-50, 96-209

☐ Further documents are listed in the continuation of Box C.

See patent family annex.

**\* Special categories of cited documents:**

- "A" document defining the general state of the art which is not considered to be of particular relevance
- "E" earlier application or patent published on or after the international filing date
- "L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)
- "O" document referring to an oral disclosure, use, exhibition or other means
- "P" document published prior to the international filing date but later than the priority date claimed

"T"

later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X"

document of particular relevance: the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

"Y"

document of particular relevance: the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art

"&amp;"

document member of the same patent family

Date of the actual completion of the international search

14 January 2002 (14.01.2002)

Date of mailing of the international search report

15 MAR 2002

Name and mailing address of the ISA/US

Commissioner of Patents and Trademarks  
 Box PCT  
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Facsimile No. (703)305-3230

Authorized officer

Dorthea Lawrence For  
Sandra Saucier

Telephone No. (703) 308-0196

# INTERNATIONAL SEARCH REPORT

International application No.

PCT/US01/15150

## Box I Observations where certain claims were found unsearchable (Continuation of Item 1 of first sheet)

This international report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. ☐ Claim Nos.:  
because they relate to subject matter not required to be searched by this Authority, namely:
2. ☐ Claim Nos.:  
because they relate to parts of the international application that do not comply with the prescribed requirements to such an extent that no meaningful international search can be carried out, specifically:
3. ☐ Claim Nos.:  
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

## Box II Observations where unity of invention is lacking (Continuation of Item 2 of first sheet)

This International Searching Authority found multiple inventions in this international application, as follows:  
Please See Continuation Sheet

1. ☐ As all required additional search fees were timely paid by the applicant, this international search report covers all searchable claims.
2. ☐ As all searchable claims could be searched without effort justifying an additional fee, this Authority did not invite payment of any additional fee.
3. ☒ As only some of the required additional search fees were timely paid by the applicant, this international search report covers only those claims for which fees were paid, specifically claims Nos.: 1-50, 96-209
4. ☐ No required additional search fees were timely paid by the applicant. Consequently, this international search report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:

Remark on Protest



The additional search fees were accompanied by the applicant's protest.



No protest accompanied the payment of additional search fees.

## INTERNATIONAL SEARCH REPORT

International application No.

PCT/US01/15150

### BOX II. OBSERVATIONS WHERE UNITY OF INVENTION IS LACKING

This application contains the following inventions or groups of inventions which are not so linked as to form a single general inventive concept under PCT Rule 13.1. In order for all inventions to be examined, the appropriate additional examination fees must be paid.

Group I, claims 1-31, drawn to a method of isolating X and Y sperm cells by determining a sex differentiation characteristic and separating on the basis of that characteristic.

Group II, claims 32-50, drawn to an apparatus to isolate X and Y sperm cells by determining a difference in volume of the sperm heads, comprising a beam of electromagnetic radiation, detector, analyzer and sperm cells.

Group III, claims 51-69, drawn to a method of separating X and Y sperm cells by determining the volume of the capsule containing the DNA.

Group IV, claims 70-95, drawn to a method of differentiating particles.

Group V, claims 96-120, drawn to an apparatus to differentiate particles comprising an asymmetric particle in a fluid stream, an irradiation source, optics, light emission material, detector and analyzer.

Group VI, claims 121-147, drawn to an apparatus to differentiate particles comprising a nozzle, an oscillator, droplets and sperm cells.

Group VII, claims 148-182, drawn to an apparatus comprising particles, light emission source, a photo-multiplier tube and an analyzer.

Group VIII, claims 183-209, drawn to an apparatus comprising asymmetric particles, an irradiation source, optics, light emission material, detector.

The inventions listed as Groups I-VIII do not relate to a single general inventive concept under PCT Rule 13.1 because, under PCT Rule 13.2, they lack the same or corresponding special technical features for the following reasons:

PCT Rules 13.1 or 13.2 do not provide for multiple methods or multiple apparatuses.

The methods of Group I, III and IV all require distinct materials, for example, the method of Group II requires assessment of the volume of the capsule containing DNA, while the methods of Groups I or IV do not require such as determination.

The methods of Groups I, III or IV do not require the apparatuses of Groups II, V, VI, VII or VIII.

The apparatuses of Groups II, V, VI, VIII all have distinct components and are, therefore, distinct apparatuses.